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A MARKED CONTRAST BETWEEN SEROTONERGIC AND DOPAMINERGIC CHANGES IN DOPA-RESPONSIVE DYSTONIA

Dopa-responsive dystonia (DRD) is a clinical syndrome characterized by childhood-onset dystonia and a dramatic and sustained response to low doses of levodopa.¹ Autosomal dominant DRD is caused by mutations in *GCH1*, which encodes GTP cyclohydrolase 1 (GTPCH), the rate-limiting enzyme in the biosynthesis of tetrahydrobiopterin (BH4; the cofactor for phenylalanine and tyrosine and tryptophan hydroxylases [TH and TPH]).¹ Recently, the phenotype of this major form of DRD (GTPCH-deficient DRD) was expanded to include nonmotor symptoms, such as depression, anxiety, and obsessive-compulsive disorder, and these psychiatric symptoms were attributed to serotonin deficiency.^{2,3} However, the actual status of brain serotonergic involvement in GTPCH-deficient DRD is unknown. To evaluate possible serotonin deficiency, we measured levels of serotonin markers in the striatum (one of the serotonin- and dopamine-rich brain regions)⁴ in a 19-year-old woman with DRD and a *GCH1* nonsense mutation.⁵ Her dystonia (onset at 5 years of age) was well controlled by levodopa (without a decarboxylase inhibitor) 750 mg/d for 11 years (no psychiatric symptoms and residual motor signs) until her automobile accidental death. Neuropathologic studies revealed a normal population of cells with reduced melanin in the substantia nigra and also no evidence of degeneration in other brain areas.^{1,5} This study was approved by the institutional review board of the Centre for Addiction and Mental Health.

Results. In the putamen and caudate nucleus of the patient with GTPCH-deficient DRD, protein concentrations of serotonin transporter, TPH, and “control” neuron-specific enolase were within the age-matched normal control range (table). Normal dopamine transporter, dopa decarboxylase protein, and vesicular monoamine transporter 2 levels and severely reduced TH protein content in the putamen were reported previously in this patient.⁵ In the striatum of the patient, we found normal concentrations of serotonin and confirmed very low levels of dopamine. The most remarkable contrast between serotonergic and dopaminergic findings in this patient was preserved TPH protein and serotonin

concentrations (118% and 79% of the age-matched normal control means) vs severely decreased TH protein and dopamine levels (less than 1.5%⁵ and 8%) in the putamen.

Discussion. Biochemical findings in GTPCH-deficient DRD have suggested that severe striatal dopamine reduction (confirmed in the present study) is caused not only by decreased TH activity due to low cofactor content but also by actual loss of TH protein without nerve terminal loss. No dopaminergic terminal reduction was indicated by the normal striatal dopamine transporter, dopa decarboxylase protein, and vesicular monoamine transporter 2 levels.^{1,5} In this autosomal dominant DRD, as BH4 is also the cofactor for TPH, it has been assumed that partial BH4 deficiency results in lowering of serotonin and that there is an increased frequency of nonmotor symptoms relating to a deficit in serotonin.^{2,3} However, the present investigation demonstrated that all of the serotonin markers in the striatum were within the control range; we cannot exclude a possibility that even low-dose levodopa treatment might have influenced serotonergic activities (but this influence should be small). Our neurochemical data suggest that the different susceptibility to brain BH4 depletion between TH and TPH could be explained by a difference in the degree of regulatory effects of BH4 on the steady-state level (stability/expression)^{1,5} of these 2 hydroxylase proteins. The human data are supported by findings of decreased TH protein but preserved TPH protein in the brain of BH4-deficient mice.⁶ Other possible reasons are different *GCH1* expression levels in dopaminergic and serotonergic neurons and differences in K_m values of the 3 hydroxylases for BH4.⁷ Our normal brain data of the serotonin markers suggest that a frequency of psychiatric symptoms in patients with GTPCH-deficient DRD on levodopa therapy might not be higher than that in a normal population. Possible loss of brain noradrenaline, which may contribute to mood changes, could be compensated by adequate treatment with levodopa. However, we cannot exclude a possibility that, when combined with additional trigger factors, a latent TPH hypofunction in GTPCH-deficient DRD might induce psychiatric symptoms more easily in patients with this disorder than in normal subjects. Nevertheless, Brüggemann et al.² have recently reported

Table SERT, TPH, and NSE protein levels and serotonin and dopamine levels in the striatum of a patient with GTPCH-deficient DRD and of normal control subjects

Variable	No. of cases	Putamen	Caudate nucleus
SERT protein			
Patient	1	0.77	0.31
Controls	9 ^a	0.87 ± 0.15 (0.39-1.63)	0.46 ± 0.07 (0.25-0.88)
TPH protein			
Patient	1	0.53	0.08
Controls	9 ^a	0.45 ± 0.09 (0.13-0.92)	0.16 ± 0.05 (0.04-0.56)
NSE protein			
Patient	1	0.82	0.74
Controls	9 ^a	0.68 ± 0.05 (0.47-0.94)	0.75 ± 0.03 (0.59-0.86)
Serotonin			
Patient	1	0.19	0.23
Controls	6 ^b	0.24 ± 0.04 (0.09-0.32)	0.30 ± 0.10 (0.08-0.75)
Dopamine			
Patient	1	0.56	1.19
Controls	6 ^b	7.07 ± 0.77 (5.34-10.71)	6.76 ± 0.66 (5.44-9.66)

Abbreviations: DRD = dopa-responsive dystonia; GTPCH = GTP cyclohydrolase 1; NSE = neuron-specific enolase; SERT = serotonin transporter; TPH = tryptophan hydroxylase.

Data are given as mean ± SE (range) in µg tissue standard protein/µg tissue sample protein for SERT, TPH, and NSE and in ng/mg of wet tissue for serotonin and dopamine. SERT, TPH, and NSE (a control) protein levels were measured by quantitative immunoblotting, and serotonin and dopamine concentrations were determined by high performance liquid chromatography-electrochemical detection.⁴ The age and postmortem time of the genetically confirmed female patient with GTPCH-deficient DRD were 19 years and 16 hours, respectively.⁵

^a For SERT, TPH, and NSE protein measurements, the ages and postmortem times of the 9 normal control subjects (9 men) were 28 ± 3 (18-38) years and 15 ± 2 (5-25) hours (mean ± SE [range]).

^b For serotonin and dopamine measurements, the ages and postmortem times of the 6 normal control subjects (4 men and 2 women) were 24 ± 3 (14-34) years and 13 ± 2 (5-22) hours.

that high frequencies of depression and anxiety found previously by their group were not confirmed in a case-control investigation using levodopa-treated patients with GTPCH-deficient DRD.

Although our findings in a single patient require replication in a representative number of cases, normal striatal TPH protein and serotonin concentrations, which are in marked contrast to substantially reduced TH protein and dopamine levels in the striatum (especially in the putamen), may contribute to characteristic symptoms (e.g., no developmental psychomotor delay) in GTPCH-deficient DRD and could distinguish this dominantly inherited GTPCH deficiency from autosomal recessive BH4-related enzyme deficiencies (for which administration of both levodopa and 5-hydroxytryptophan is necessary

because of dopamine and serotonin deficits),¹ including recessively inherited GTPCH deficiency.

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